A Cytotaxonomic Study of Five Species of *Impatiens* (Balsaminaceae) in Java and Borneo, Malesia

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Somatic chromosome numbers of five species belonging to the genus *Impatiens* (Balsaminaceae) collected from Java and Borneo, Malesia are reported. *Impatiens chonoceras* Hassk. was found to be 2n = 14, *I. javensis* Steud., *I. platypetala* Lindl., and *I. radicans* Zoll. were 2n = 16, and *I. kinabaluensis* S. Akiyama & H. Ohba was 2n = 12. Chromosome numbers of *I. chonoceras*, *I. javensis*, *I. radicans*, and *I. kinabaluensis* were examined for the first time. Karyotypes of *I. javensis*, *I. platypetala*, and *I. radicans* are similar and are thought to be closely related to each other. Taxonomical notes for each species are given.

Key words: Chromosome, cytotaxonomy, Impatiens, karyomorphology, Malesia.

Impatiens (Balsaminaceae) consists of 450 to 850 species distributed in tropical and temperate regions in the northern hemisphere (Grey-Wilson 1980, Cronquist 1981). Southeast Asia including Malesian region is one of the centers of diversity for *Impatiens* (Grey-Wilson 1980).

Chromosome numbers of *Impatiens* have been examined by many authors and various somatic chromosome numbers, 2n = 6, 8, 12, 14, 15, 16, 17, 18, 19, 20, 26, 28, 32, 34, 36, 40, 44, 48, 50, 56, ca. 60, have been reported (see Federov 1969, Goldblatt 1981, 1984, 1985, 1988, Goldblatt and Johnson 1990, 1991, 1994, 1996, 1998, 2000, 2003). Thus

polyploidy and aneuploidy with different basic chromosome numbers might play an important role in the speciation of *Impatiens*.

Karyological studies have been made for species of *Impatiens* in several regions. For example, India (Govindarajan and Subramanian 1986, Rao et al. 1986), the Himalayas (Akiyama et al. 1992), and Southwest China (Sugawara et al. 1994, 1997). For Southeast Asian species of *Impatiens*, although several chromosome counts have been reported (Jones and Smith 1966, Arisumi 1973, 1987, Shimizu 1979, Larsen 1981, Okada 1989), no karyological study has been done.

Table 1. Species examined, localities and chromosome numbers of five species of Impatiens

Species	Localities (voucher specimens and their deposition)	Chromosome number (2n)
I. chonoceras Hassk.	Indonesia. West Java (Jawa Barat); Mt. (Gn.) Papandajan, 1950–2470 m alt. (Tsukaya & al. 104, 9 July 2003, TNS, BO)	14*
I. javensis Steud.	Indonesia. West Java (Jawa Barat): Gn. Gede-Pangorango National Park, 1840 m alt. (Tsukaya & al. 235, 6 March 2003, TNS BO)	16*
I. platypetala Lindl.	Indonesia. West Java (Jawa Barat); Gn. Gede-Pangorango National Park, 2120 & 1840 m alt. (Tsukaya & al. 259, 260, 8 March 2003, TNS, BO)	16
I. radicans Zoll.	Indonesia. West Java (Jawa Barat): Gn. Gede-Pangorango National Park, Gn. Gede/Gn. Pangorango, 2400 m alt. (Tsukaya & al. 240, 7 March 2003, TNS, BO)	16*
I. kinabaluensis S. Akiyama & H. Ohba	Malaysia. Sabah (Borneo Island); Kinabalu National Park, Mt. Kinabalu, Timpohon Gate-Layang-Layang, 1770 m alt. (Tsukaya & al. 0403201, 14 March 2004, TNS, BORH**)	12*

^{*}New count for the taxon.

This paper reports the chromosome numbers and karyotypes of five species of *Impatiens* collected from Java and Borneo, Malesia.

Materials and Methods

Materials were collected from their native habitat in West Java (Indonesia) and Mt. Kinabalu, Borneo (Sabah, Malaysia) in 2003 and 2004. Localities and voucher specimens are listed in Table 1.

For observation of somatic metaphase chromosomes, root tips were pretreated in the field with a 2 mM 8-hydroxyquinoline solution for about 3 hours and fixed with Newcomer's fluid (see Sharma and Sharma 1980, Wakabayashi 1988). The root tips were stained by Feulgen's nuclear reaction, and macerated in a mixture of 2 % pectinase and 2 % cellulase for 1.5–2 hours. They were again stained with 2 % lacto-propionic orcein and then squashed and observed using a light microscope.

Results and Discussion

Chromosome number of I. chonoceras

was 2n = 14, *I. javensis*, *I. platypetala*, and *I. radicans* were 2n = 16, and *I. kinabaluensis* was 2n = 12, respectively (Table 1). Among them, chromosome numbers of *I. chonoceras*, *I. javensis*, *I. radicans*, and *I. kinabaluensis* were counted for the first time.

1. Impatiens chonoceras Hassk. (Fig. 1a)

The somatic chromosome number of I. chonoceras was 2n = 14. This number is considered to be diploid, with basic chromosome number x = 7. Chromosomes are $1.5-3.1~\mu m$ long. Karyologically, the complement is monomodal, gradually reducing in size. Satellites were observed in two pairs of chromosomes: in the longest and second longest pairs.

Akiyama et al. (1992) reported chromosome numbers of 16 species in the Himalayas and reported those of *I. falcifer* and *I. serrata* as 2n = 14. Sugawara et al. (1997) reported 15 species of *Impatiens* in Yunnan, SW China and chromosome numbers of *I. kamtilongensis* Toppin and *I. wuchengyihii* S. Akiyama, H. Ohba & S. K. Wu as 2n = 14. All the species mentioned

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above are similar in having characteristic floral morphology; the shape of the lower sepal is navicular tapering to the spur and the inflorescence usually has one or two flowers (Akiyama et al. 1991, 1996). Morphologically I. chonoceras is similar to these species. Impatiens chonoceras has a lower sepal with pubescence on the outer surface, like I. kamtilongensis and I. wuchengyihii, while the lower sepals of I. falcifer and I. serrata are glabrous on the outer surface. The inflorescence of I. chonoceras is axillary and pedunculate, and usually consists of two flowers with a basal bract. These two flowers appear in opposite. The inflorescences of I. kamtilongensis and I. wuchengyihii are racemose with two flowers (Akiyama et al. 1996), while those of *I. falcifer* and *I. serrata* are unique and have a middle bract (Akiyama and Ohba 2000). It is notable that I. chonoceras has the apparently most primitive form of inflorescence among Chinese species (i. e., I. kamtilongensis and I. wuchengyihii) and Himalayan species (i. e., I. falcifer and I. serrata).

Cytologically, all the species mentioned above have similar karyotypes. *Impatiens chonoceras*, *I. kamtilongensis*, and *I. wuchengyihii* have somatic chromosome numbers 2n = 14, with monomodal complements. *Impatiens falcifer* and *I. serrata* also have somatic metaphase chromosomes 2n = 14, and they show monomodal complements as judged from the plates (Akiyama et al. 1992).

Impatiens chonoceras was collected in Mt. Papandajan, a famous volcano located in SW Java. From the morphology and cytology, I. chonoceras is considered to be closely related to the species of the Himalayas and adjacent regions. Potentilla polyphylla Wall. ex Lehm. (Rosaceae), distributed mainly in the Himalayas and adjacent regions, was collected in this mountain (Kalkman 1968, 1993), suggesting that Mt. Papandajan had been a refugium for Himalayan plants in this

area.

2. *Impatiens javensis* Steud., *I. platypetala* Lindl. and *I. radicans* Zoll. (Figs. 1b–e)

The somatic chromosome numbers of I. javensis, I. platypetala, and I. radicans were 2n = 16. These numbers are considered to be diploid, with basic chromosome number x = 8. Chromosomes are $0.9-1.8 \mu m$ long in I. javensis, $1.0-1.5 \mu m$ in I. platypetala, and $1.1-1.7 \mu m$ in I. radicans. Karyomorphologically they showed similar karyotypes. The complements were monomodal, gradually reducing in size. Satellites were observed in one pair of chromosomes.

Van Steenis (1948) recognized three subspecies in I. platypetala: subsps. platypetala, aurantiaca, and nematoceras. Among the subspecies, subsp. platypetala has the widest distribution range, throughout Java and Sumatra. Zinov'eva-Stahevitch and Grant (1984) reported chromosome numbers of *I*. platypetala subsp. platypetala as n = 7 and 2n = 14 (from Mt. Gedé, Java), subsp. aurantiaca 2n = 14 (from Sulawesi), and subsp. nematoceras n = 8 and 2n = 16 (from Indonesia). Arisumi (1987) reported the chromosome number of I. platypetala (subsp.) aurantiaca as 2n = 8. Okada (1989) counted chromosome numbers platypetala from Sumatra, which may be subsp. platypetala based on distribution, as 2n = 16. Our count for *I. platypetala*, refers to subsp. platypetala, coincides with the number counted by Okada (1989), but differs from the number given by Zinov'eva-Stahevitch and Grant (1984) for a collection from the same locality. For confirming the chromosome numbers of the species and the infraspecific taxa of I. platypetala, examination of ample materials collected from the entire range of distribution is necessary.

Impatiens platypetala, I. javensis, and I. radicans are closely related species judging from floral morphology and karyotypes. In Gunung (Mt.) Gede-Pangrango National

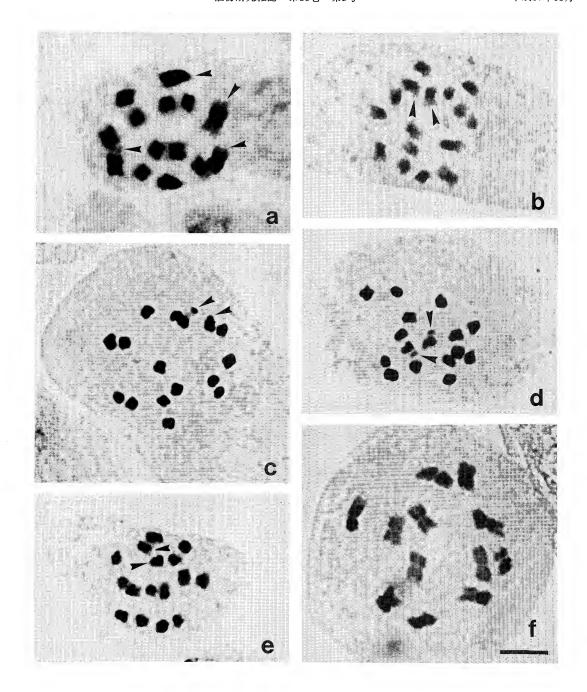


Fig. 1. Somatic metaphase chromosomes of five species of *Impatiens* in SE Asia. a: *I. chonoceras* (2n = 14). b: *I. javensis* (2n = 16). c, d: *I. platypetala* (2n = 16). e: *I. radicans* (2n = 16). f: *I. kinabaluensis* (2n = 12). Arrows indicate satellite chromosomes. Bar indicates 5 μ m.

Park, they inhabit different altitudes: I. platypetala at the lowest altitudes, less than 1800 m, I. javensis between 1800 m and 2400 m, and I. radicans above 2400 m. Impatiens platypetala is distributed widely in Malesia while I. javensis is found in Java and probably in Sumatra, and I. radicans is endemic to West Java (as far as known, from Mts. Pangrango, Tjikurai and Papandajan) (van Steenis 1972). Impatiens javensis and I. radicans are genetically close and it is known that they produce interspecific hybrid (Tsukaya 2004). It may be thought that I. javensis and I. radicans might be differentiated in diploid level after altitudinal isolation from an I. platypetala-like ancestor having 16 somatic chromosomes in Java Island.

3. *Impatiens kinabaluensis* S. Akiyama & H. Ohba (Fig. 1f)

The somatic chromosome number of I. kinabaluensis was 2n = 12. The number is considered to be diploid, with basic chromosome number x = 6. Chromosomes are 2.4-3.8 µm long. Karyologically, the complement is monomodal, gradually reduced in size. Satellites were not observed.

Chromosome numbers of Impatiens in Southeast Asia including Malesia have been fragmentarily reported (Jones and Smith 1966, Arisumi 1973, 1987, Shimizu 1979, Larsen 1981, Zinov'eva-Stahevitch and Grant 1984, Okada 1989), with those of about 20 species so far revealed. Among these species, two species were reported as 2n = 12; I. chiangdaoensis T. Shimizu from Thailand (Shimizu 1979, Larsen 1981) and "I. harlandii Dransfield ined." from Borneo (Zinov'eva-Stahevitch and Grant 1984) although we could not find any literature on "I. harlandii", and could not determine whether this is the same species as I. kinabaluensis or not.

Van Steenis (1948) noted the distribution of *I. platypetala* subsp. *platypetala* as "throughout Java, further in Sumatra and

probably also occurs in Borneo, Sulawesi and the Lesser Sunda Islands". Masamune (1942) listed *I. platypetala* among Bornean *Impatiens*.

After examining the morphology of *Impatiens* collected in Mt. Kinabalu, Borneo (Tsukaya & al. 0403201), it is concluded that this is not *I. platypetala*, but a new species, named *I. kinabaluensis* (Akiyama et al. 2005)

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池田 博*, 秋山 忍*, 塚谷裕一*, M. マハメド*, D. ダルナエディ*: ジャワ島およびボルネオ島産ツリフネソウ属植物(ツリフネソウ科)の細胞分類学的研究

ジャワ島 (インドネシア) およびボルネオ島キナバル山 (マレーシア) で採集されたツリフネソウ科ツリフネソウ属 5種の染色体数と核型を報告した.

Impatiens chonoceras Hassk.は 2n = 14, I. javensis Steud., I. platypetala Lindl., I. radicans Zoll.は 2n = 16, I. kinabaluensis S. Akiyama & H. Ohba は 2n = 12 であった. Impatiens chonoceras, I. javensis, I. radicans, I. kinabaluensis の染色体数は、今回が初めての報告である. Impatiens chonoceras は、花や花序の形態と核型から、ヒマラヤや中国南西部に分布する種と類縁があると考えられた. Impatiens javensis, I. platypetala, I. radicans の 3 種は、花の形態と核型から互いに近縁と考えられ、分布がジャ

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